STUDIES IN MICROSCOPY OF THE SURFACE OF THE SKIN

Preliminary Report of Technics [1, 2]

LEON GOLDMAN, M.D. and WALDO YOUNKER, Ch.E.

Examination of the cutaneous surface by means of compound microscopes has been neglected, for the most part, in clinical dermatology. This is indeed surprising since such observations, initiated more than a year ago in the search for cutaneous filariae (1) by direct skin microscopy, have given some interesting and practical results.

Simple magnifiers are used commonly in modern dermatologic practice. These magnifiers consist of single or multiple lens magnifiers, with powers of magnification ranging from approximately 3 × to a maximum of 20 ×. However, magnification by this means above 10 × is impractical. Head loupes, which afford stereoptic vision, give only about threefold magnification. A recent development of the simple magnifier has been the illuminated type. It is our opinion that the simple magnifier is much more impressive to the patient than it is valuable to the observer. Distortion, focal length for high magnification, chromatic aberration, (corrected for in the achromatic magnifier) are some of the properties which tend to limit the value of this instrument in clinical dermatology.

The compound microscope gives better definition and resolution for examination of the cutaneous surface of the patient. The initial interest in skin microscopy appears to have developed in relation to capillaroscopy some years ago (2), but was not maintained by clinical dermatologists possibly because of the lack of practical value from such observations. R. and F. Jaeger (3, 4) and Schmidt-La Baume (5) have revived interest in direct examination of the skin by the use of the compound microscope; they employed the binocular microscopes for the lower powers and the Ultrapak for studies with higher magnifications. In this microscope the light enters the microscope from the side and is reflected down on the object. R. and F. Jaeger (3, 4) used objectives UO65 and UO11 and oculars 8–10 ×. To increase the value of definition of the cutaneous surface, these observers used primulin and auramine as fluorochromes directly on the skin. Fluorescent microscopy of the skin surface was of value, for example, in examining the cutaneous surface of persons afflicted with occupational dermatoses. Using an indirect technic, Wolf (6) made celloidion or celluloid-acetone casts of the cutaneous surface and studied these with high power magnifications. The adhesion specimens were fixed on a glass slide.

[1] From the Department of Dermatology and Syphilology and the Kettering Laboratory of Applied Physiology of the College of Medicine, of the University of Cincinnati.

[2] Scientific Equipment furnished by W. B. Rayton and L. V. Foster of the Scientific Bureau of Bausch and Lomb Optical Company.

Received for Publication 10/3/46

We have used the following forms of compound microscopes for examination of the skin:

1. Brinell microscope
2. Greenough type binocular microscope
3. High-power biologic microscope
4. Shop microscope
5. Vertical illuminators

The Brinell microscope is used chiefly in mechanical industries. This microscope is used to measure diameters of impressions made by the ball of a Brinell hardness testing machine. This microscope lends itself readily to examination

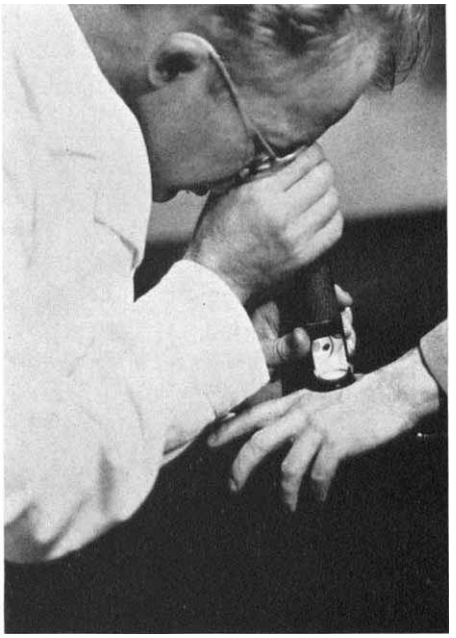

FIG. 1. PICTURE SHOWING THE USE OF THE BRINELL MICROSCOPE

of the surface of the skin including capillaroscopy. Magnifications up to 40 × may be secured with the newer models of this type and we have worked from 12.7 × to 40 ×. The light source has been either a self contained battery unit or daylight, or ultraviolet light from a lamp equipped with a Woods filter. The Shop microscope, which we have used also, can give magnifications of 40–50 ×. This Shop microscope type is essentially a portable microscope tube with a self contained battery illuminator source which illuminates from the side. With this arrangement, surface examinations are possible. In the Brinell, the illumination is chiefly vertical, in the Shop microscope, the illumination is chiefly by rays of low obliquity. When fluorescent light is employed in mocroscopy operator must guard against injury to his own eyes and those of the patient. This is especially true at present under conditions involving the use of the

ordinary sources of fluorescent light in view of the intensity required for satisfactory observation. A simple and properly designed microscope fluorescence lamp or light concentration apparatus would serve to eliminate some of these difficulties. The scale of the Brinell microscope is often a minor liability in that it covers some parts of the lesion, and makes it impossible to obtain accurate measurement of the entire cutaneous lesion. The other types of compound microscopes have been less flexible in use in our hands and have given more difficulty from the aspects of manipulation and illumination. The Greenough binocular microscope gives good definition, high magnifications, up to 150 ×, but does not give the resolution and detail that the high power biologic microscope

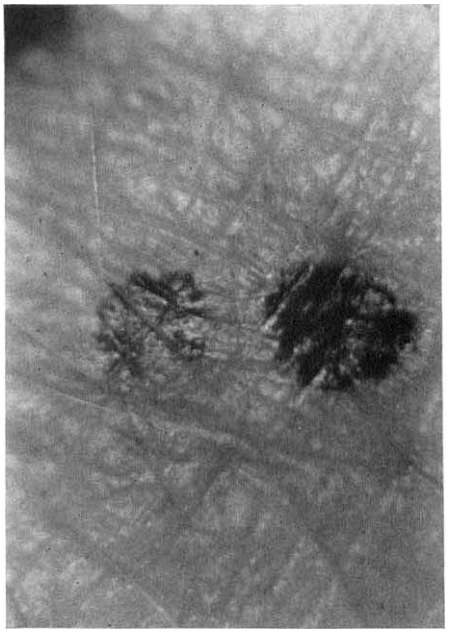

FIG. 2. PHOTOGRAPH TAKEN OF SKIN SURFACE, WITH SILVER NITRATE SPOTTING, THROUGH BRINELL MICROSCOPE × 20

can offer. Of the vertical illuminators the dark field vertical illuminator type is the most efficient.

Illumination of the skin for surface microscopy is important and should provide good vertical, oblique, "silhouette" illumination or transillumination according to the requirement. Briefly, examination of the skin surface itself demands vertical or oblique or "silhouette" lighting with little light from deeper tissue inside the skin. Capillaroscopy demands elimination of almost all the reflected glare from the surface of the skin and flattening of the surface. Examination for animal parasites requires transillumination of the cutaneous area. The illumination source which we have used has been chiefly the Nicholas illuminator, with modification. The illuminator is equipped with a condensing system which projects a beam of light. This apparatus offers high intensity

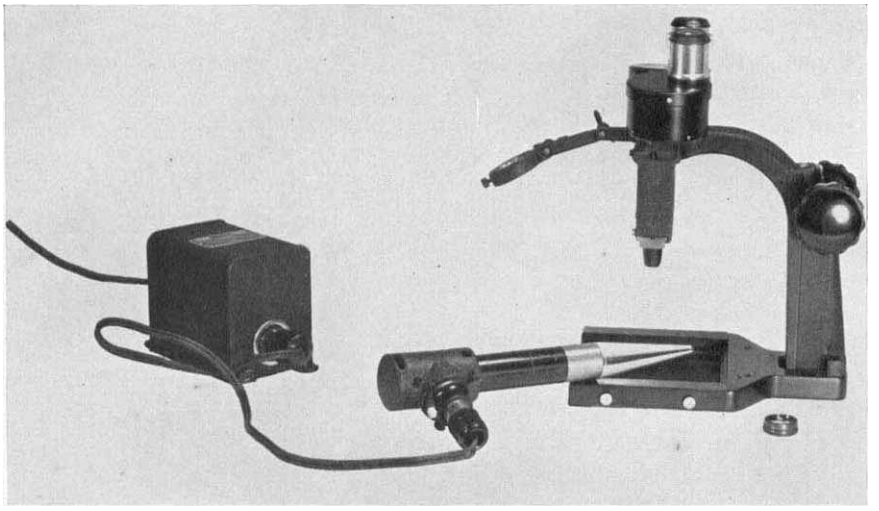

FIG. 3. GREENOUGH TYPE BINOCULAR MICROSCOPE WITH NICHOLAS ILLUMINATOR WITH LUCITE ROD ATTACHMENT FOR VARIED TYPES OF ILLUMINATION OF SURFACE OF SKIN

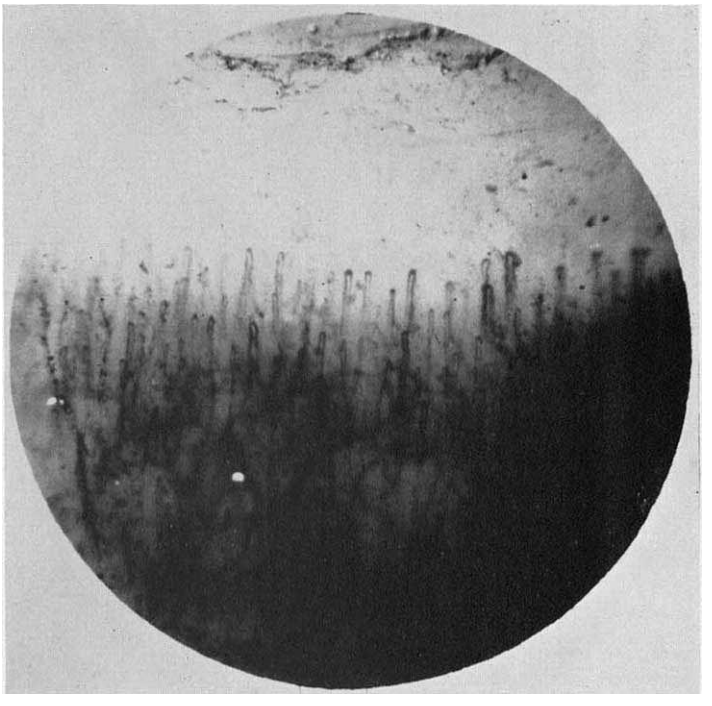

FIG. 4. CAPILLARIES BASE OF FINGER NAILS, BIOLOGICAL MICROSCOPE, 12.5 × EYEPIECE, 32 MM. OBJECTIVE, × 50 (FROM THE COLLECTION OF L. V. FOSTER.)

and can be attached to a binocular microscope. The disadvantages are the heat produced and projected on the skin and some lack of flexibility, which is so desirable in examinations of different cutaneous surfaces. To overcome these disadvantages, one of us (W. Y.) has devised a lucite attachment which remains cold, provides a small intense light spot and serves also for trans-illumination procedures. Another plastic, polystyrene, has also been suggested as an attachment for the Nicholas illuminator. At times, we have made use of microscope lamps using the 6 volt bulb type. Intense illumination is frequently necessary in microscopy of the skin, especially with higher magnifications and, of course, with photomicroscopy. Flash light is necessary for cutaneous photomicroscopy. The microscope and its illuminator comprise a heavy examination unit not easily adaptable to the examination of various areas of the skin. Attachment to a suitable type of adjustable stand will help to make this unit more flexible. In our preliminary observations color filters and polaroid discs have not been of great help. Polaroid discs may help to cut down the glare. Fluorescent microscopy has also been used especially with the Greenough type of binocular microscope. The recent developments in phase contrast microscopy suggest many practical uses for examination of the skin. Preliminary experiments with this microscope have been disappointing in detecting fungous elements in the skin and hair but additional work must be done. The recent developments in electron optics especially as related to infra-red telescopes may offer some value to clinical and investigative dermatology. The use of selective-"vital" staining and radio-active tracers is contemplated to amplify the field of skin microscopy.

In brief, our observations have included classification of the pictures of normal, dry, seborrheic, pigmented and nonpigmented types of skin. The Negro skin yields excellent pictures. We have used fluorescent microscopy to study disappearance of pigment in advancing vitiligo. The use of fluorescence microscopy has been extended with the topical applications of dilute solution of fluorescein to the surface of the skin. Auramine has also been tried. Freckles, petechiae, hemangiomas of the spider type, warts and callosities have been examined and additional studies on hemangiomas of the spider type are contemplated. Surface microscopy with and without fluorescein solutions is being used in the study of primary irritation of the skin, in so far as this is evidenced by erythema, pigmentation, desquamation and fissuring. Also the reaction of the capillary loops in normal skin and in that sensitized (Schmidt-La Baume) to materials placed upon its surface is under study. Microscopy of the structure of the "normal" skin of the neck in cases of papilloma colli has been performed in order to observe the early development of these lesions. The identification of cutaneous parasites has been attempted. As mentioned previously, a search for microfilariae of *Onchocerca volvulus* and even of *Wuchereria bancrofti* was unsuccessful. Similar unsuccessful results were obtained in the search for Demodex. Transillumination of scabetic burrows has shown the outlines of the burrow with vague spotty shadows but better resolution and detail must be obtained here for absolute differentiation. Fluorescence micros-

copy has been employed here also. Casual examination of infestation of the skin by mites of the genus Trombicula have been negative but controlled mite feeding experiments are to be carried out. Critical studies, combining cutaneous microscopy with frequent biopsies, have been started for pigmented naevi, senile keratoses and cutaneous malignancies. Fungous infections are being studied by means of fluorescence microscopy.

SUMMARY

The long-neglected study of the skin surface by microscopy should be revived in the light of newer developments in the field of optics. The commonly employed simple magnifiers are of little real practical value. The compound microscope, especially of the metallurgical type, with special lighting arrangements including fluorescent illumination offers promise in clinical and investigative dermatology. Our preliminary studies have been concerned chiefly with classification of skin types, pigmentary anomalies, early papillomatous changes, early recognition of primary irritation, fungous infections, and search for parasites.

REFERENCES

1. Goldman, Leon and Ortiz, Luis Figueroa: Types of Dermatitis in American Onchocerciasis, Arch. Dermat. & Syph. **53:** 79, (Feb.) 1946.
2. Foster, L. V.: Personal communication to the authors.
3. Jaeger, R. und F.: Fluoreszenzmikroskopie in Auffallenden Licht unter besonderer Berucksichtigung der Struktur der Oberfläche der lebenden Haut und der Vereinfachung der Hilfsmittel, Ztschr., f. wissensch. Mikr., **56:** 273, 1939.
4. Jaeger, R. und F.: Die Haut Oberflächenstruktur, Arch. f. Gewerbepath. u. Gewerbehyg., **9:** 276, 1939.
5. Schmidt-La Baume: F.: Die Bedeutung der Fluoreszenzaufllicht und Kapillar Mikroskopie fur Gewerbliche Hauterkrankugen, Dermat. Wchnschr., **110:** 81, (27 Jan.) 1940.
6. Wolf, Jan: Uber die Herstellung Mikroskopischer Präparate der Oberflächen Verschiedener Objekte mit Hilfe der Adhäsionsmethode, Ztschr. f. wissensch. Mikr., **56:** 181, 1939.
7. Goldman, Leon: A Transparency Test for the Differential Diagnosis of Plantar Warts, Arch. Dermat. & Syph., **46:** 74, (Nov.) 1942.